

SITE-SPECIFIC LABELING OF AFFINITY TAGS IN FUSION PROTEINS

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ABSTRACT

The present invention provides methods and fluorescent compounds that facilitate detecting and labeling of a fusion protein by being capable of selectively binding to an affinity tag.

The fluorescent compounds have the general formula $A(B)_n$, wherein A is a fluorophore, B is

10 a binding domain that is a charged chemical moiety, a protein or fragment thereof and n is an integer from 1-6 with the proviso that the protein or fragment thereof not be an antibody or generated from an antibody. The present invention provides specific fluorescent compounds and methods used to detect and label fusion proteins that contain a poly-

histidine affinity tag. These compounds have the general formula $A(L)_m(B)_n$ wherein A is a

15 fluorophore, L is a linker, B is an acetic acid binding domain, m is an integer from 1 to 4 and n is an integer from 1 to 6. The acetic acid groups interact directly with the positively charged histidine residues of the affinity tag to effectively label and detect a fusion protein containing such an affinity tag when present in an acidic or neutral environment.